

FIG 1A. PRODUCTION OF GP88 BY  
TUMORIGENIC AND NON-TUMORIGENIC  
CELLS

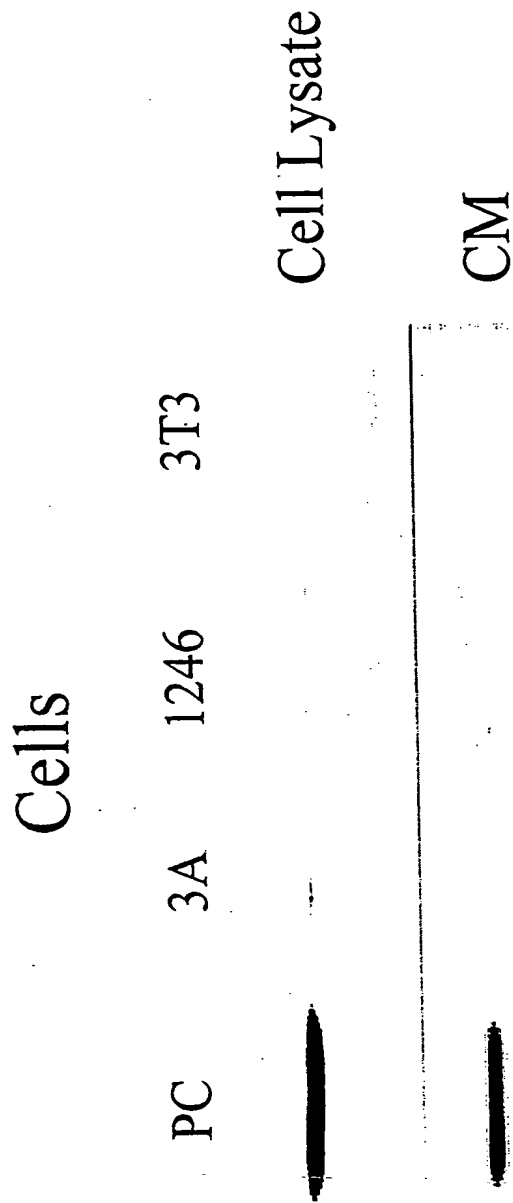


FIG 1B. GP88 mRNA EXPRESSION

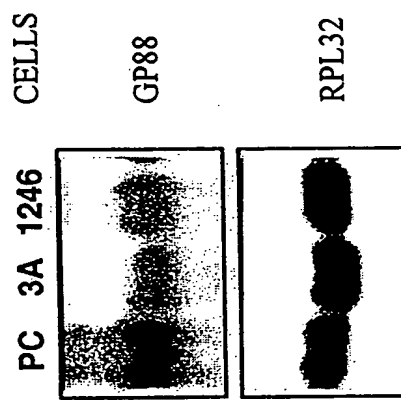
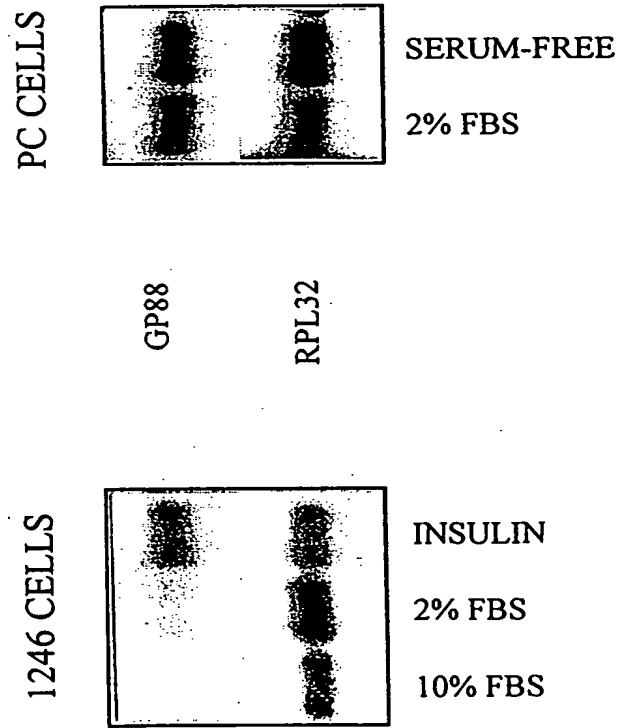


FIG 1C. GP88 mRNA EXPRESSION IN  
VARIOUS CULTURE CONDITIONS



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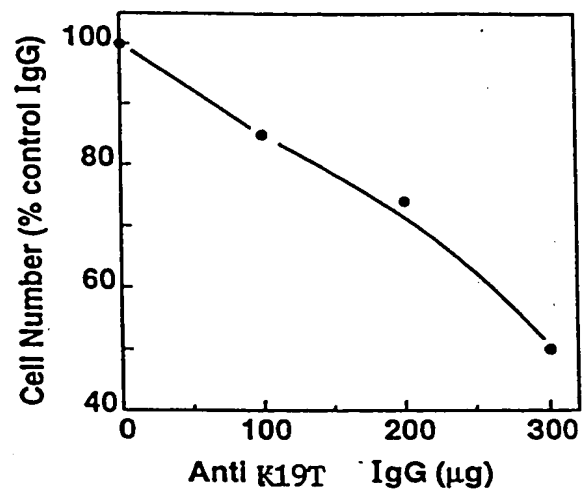
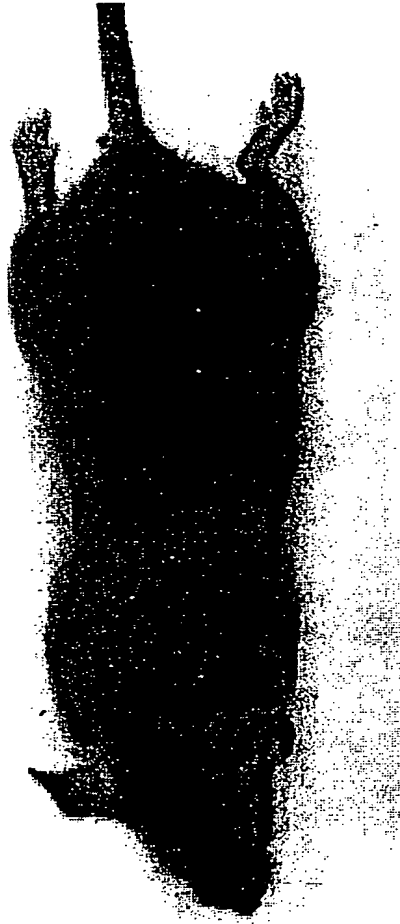


FIG. 2

FIG 3. ABSENCE OF TUMOR FORMATION IN C3H MICE BY INHIBITION OF GP88  
EXPRESSION



GP88 ANTISENSE TRANSFECTED PC CELLS



CONTROL TRANSFECTED PC CELLS

FIG 4. GP88 PROTEIN EXPRESSION IN TUMOR  
AND SURROUNDING TISSUES

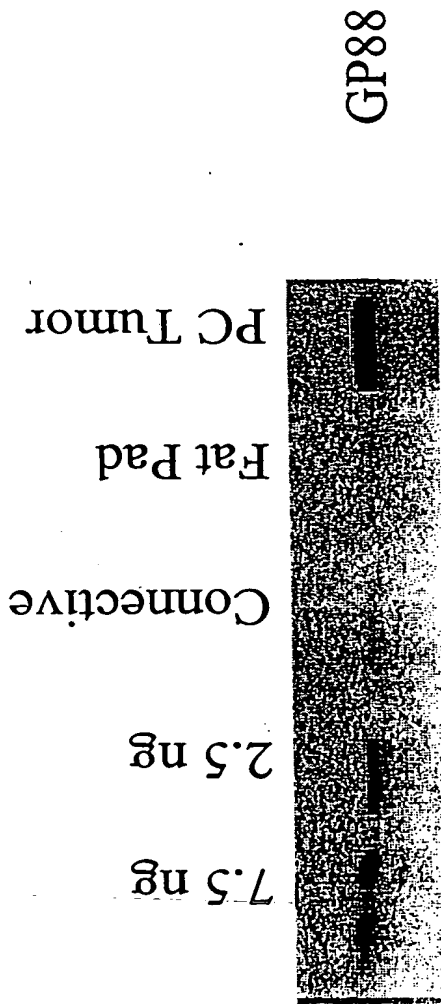
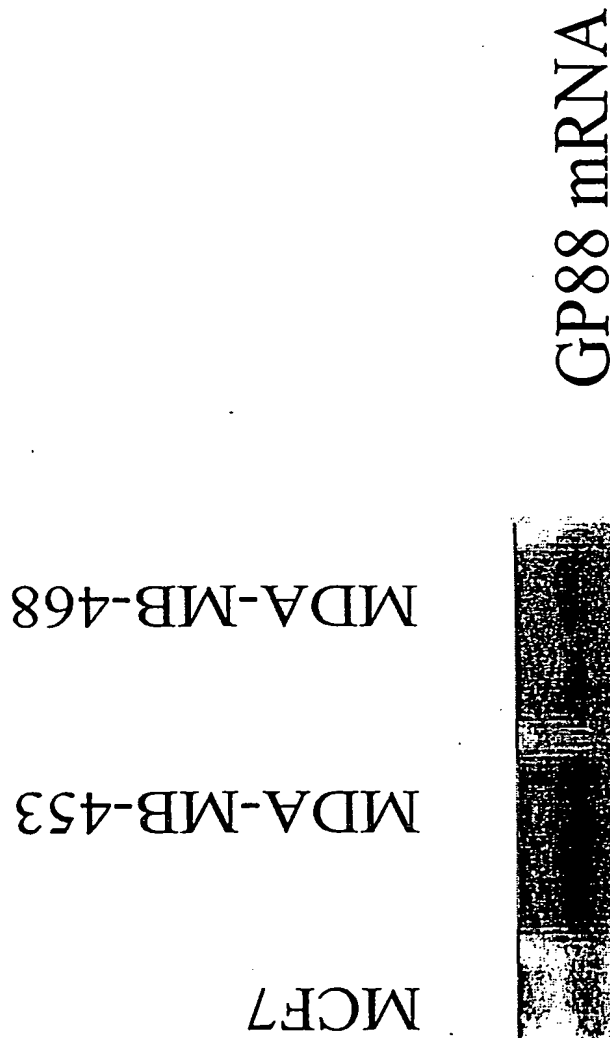


FIG 5. GP88 mRNA EXPRESSION IN  
ESTROGEN-DEPENDENT AND INDEPENDENT  
HUMAN MAMMARY CARCINOMA CELLS



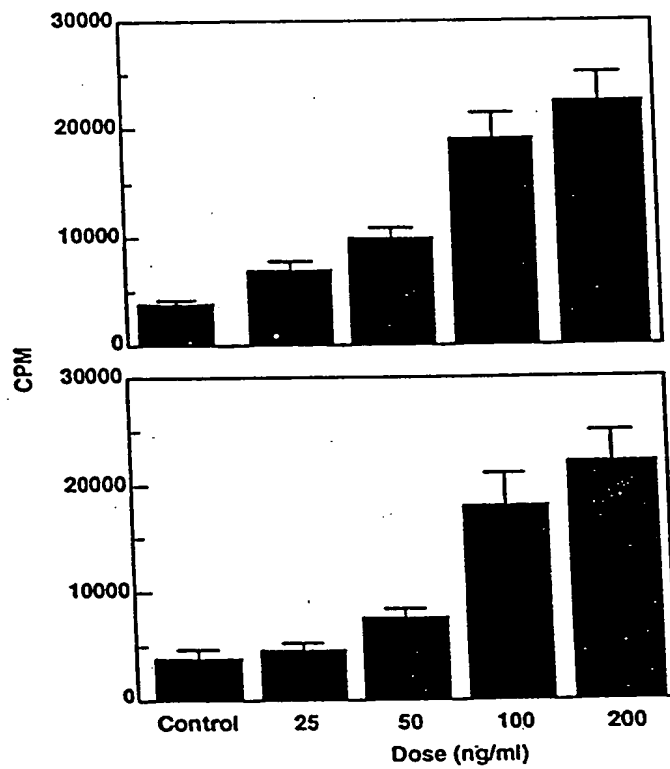
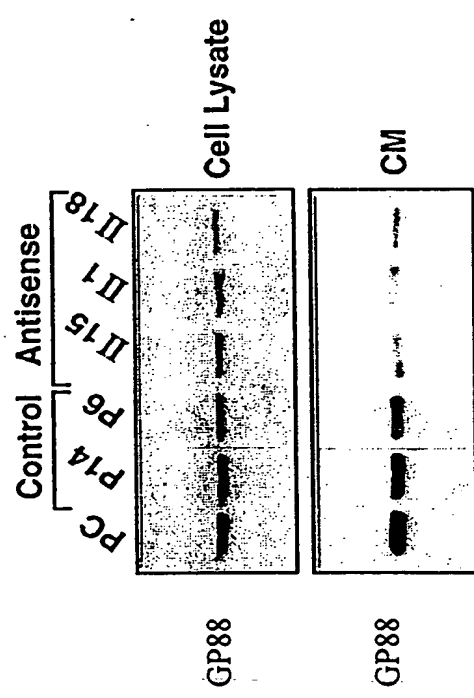


FIG. 6

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FIG 7. EXPRESSION OF GP88 IN ANTISENSE AND  
CONTROL TRANSFECTED PC CELLS



[C	GGA	CCC	CGA	CSC	AGA	CAG	ACC	ATG	TGG	GTC	CTG	ATG	AGC	TGG	CTG	46
								M	W	V	L	M	S	W	L	8
GCC	TTC	GCG	GCA	GGG	CTG	GTA	GCC	GGA	ACA	CAG	TGT	CCA	GAT	GGG	CAG	94
A	F	A	A	G	L	V	A	G	T	Q	C	P	D	G	Q	24
TTC	TGC	CCT	GTT	GCC	TGC	TGC	CTT	GAC	CAG	GGA	GGA	GCC	AAC	TAC	AGC	142
F	C	P	V	A	C	C	L	D	Q	G	G	A	N	Y	S	40
TGC	TGT	AAC	CCT	CTT	CTG	GAC	ACA	TGG	CCT	AGA	ATA	ACG	AGC	CAT	CAT	190
C	C	N	P	L	L	D	T	W	P	R	I	T	S	H	H	56
CTA	GAT	GGC	TCC	TGC	CAG	ACC	CAT	GGC	CAC	TGT	CCT	GCT	GGC	TAT	TCT	238
L	D	G	S	C	Q	T	H	G	H	C	P	A	G	Y	S	72
TGT	CTT	CTC	ACT	GTG	TCT	GGG	ACT	TCC	AGC	TGC	TGC	CCG	TTC	TCT	AAG	286
C	L	L	T	V	S	G	T	S	S	C	C	P	F	S	K	88
GGT	GTG	TCT	TGT	GGT	GAT	GGC	TAC	CAC	TGC	TGC	CCC	CAG	GGC	TTC	CAC	334
G	V	S	C	G	D	G	Y	H	C	C	P	Q	G	F	H	104
TGT	AGT	GCA	GAT	GGG	AAA	TCC	TGC	TTC	CAG	ATG	TCA	GAT	AAC	CCC	TTG	382
C	S	A	D	G	K	S	C	F	Q	M	S	D	N	P	L	120
GGT	GCT	GTC	CAG	TGT	CCT	GGG	AGC	CAG	TTT	GAA	TGT	CCT	GAC	TCT	GCC	430
G	A	V	Q	C	P	G	S	Q	F	E	C	P	D	S	A	136
ACC	TGC	TGC	ATT	ATG	GTT	GAT	GGT	TCG	TGG	GGA	TGT	TGT	CCC	ATG	CCC	478
T	C	C	I	M	V	D	G	S	W	G	C	C	P	M	P	152
CAG	GCC	TCT	TGC	TGT	GAA	GAC	AGA	GTG	CAT	TGC	TGT	CCC	CAT	GGG	GCC	526
Q	A	S	C	C	E	D	R	V	H	C	C	P	H	G	A	168
TCC	TGT	GAC	CTG	GTT	CAC	ACA	CGA	TGC	GTT	TCA	CCC	ACG	GGC	ACC	CAC	574
S	C	D	L	V	H	T	R	C	V	S	P	T	G	T	H	184
ACC	CTA	CTA	AAG	AAG	TTC	CCT	GCA	CAA	AAG	ACC	AAC	AGG	GCA	GTG	TCT	622
T	L	L	K	K	F	P	A	Q	K	T	N	R	A	V	S	200
TTG	CCT	TTT	TCT	GTC	GTG	TGC	CCT	GAT	GCT	AAG	ACC	CAG	TGT	CCC	GAT	670
L	P	F	S	V	V	C	P	D	A	K	T	Q	C	P	D	216
GAT	TCT	ACC	TGC	TGT	GAG	CTA	CCC	ACT	GGG	AAG	TAT	GGC	TGC	TGT	CCA	718
D	S	T	C	C	E	L	P	T	G	K	Y	G	C	C	P	232
ATG	CCC	AAT	GCC	ATC	TGC	TGT	TCC	GAC	CAC	CTG	CAC	TGC	TGC	CCC	CAG	766
M	P	N	A	I	C	G	S	D	H	L	H	C	C	P	Q	248
GAC	ACT	GTA	TGT	GAC	CTG	ATC	CAG	AGT	AAG	TGC	CTA	TCC	AAG	AAC	TAC	814
D	T	V	C	D	L	I	Q	S	K	C	L	S	K	N	Y	264
ACC	ACG	GAT	CTC	CTG	ACC	AAG	CTG	CCT	GGA	TAC	CCA	GTG	AAG	GAG	GTG	862
T	T	D	L	L	T	K	L	P	G	Y	P	V	K	E	V	280
AAG	TGC	GAC	ATG	GAG	GTG	AGC	TGC	CCT	GAA	GGA	TAT	ACC	TGC	TGC	CGC	910
K	C	D	M	E	V	S	C	P	E	G	Y	T	C	C	R	296
CTC	AAC	ACT	GGG	GCC	TGG	GGC	TGC	TGT	CCA	TTT	GCC	AAG	GCC	GTG	TGT	958
L	N	T	G	A	W	G	C	C	P	F	A	K	A	V	C	312

FIG. 8

Mouse .GP88 c (continued)

TGT	GAG	GAT	CAC	ATT	CAT	TTC	TGC	CCG	GCA	GGG	TTT	CAG	TGT	CAC	ACA	1006
C	E	D	H	I	H	C	C	P	A	G	F	Q	C	H	T	326
GAG	AAA	GGA	ACC	TTC	GAA	ATG	GGT	ATC	CTC	CAA	GTA	CCC	TGG	ATG	AAG	1054
E	K	G	T	C	E	M	G	I	L	Q	V	P	W	M	N	344
AAG	GTC	ATA	GCC	CCC	CTC	TTC	CTG	CCA	GAC	CCA	CAG	ATC	TTG	AAG	AGT	1102
K	V	I	A	P	L	R	L	P	D	P	Q	I	L	K	S	360
GAT	ACA	CCT	TGT	GAT	GAC	TTC	ACT	AGG	TGT	CCT	ACA	AAC	AAT	ACC	TGC	1150
D	T	P	C	D	D	F	T	R	C	P	T	H	H	T	C	376
TGC	AAA	CTC	AAT	TCT	GGG	GAC	TGG	GGC	TGC	TGT	CCC	ATC	CCA	GAG	GCT	1198
C	K	L	N	S	G	D	W	G	C	C	P	I	P	E	A	392
GTC	TGC	TGC	TCA	GAC	AAC	CAG	CAT	TGC	TGC	CCT	CAG	GGC	TTC	ACA	TGT	1246
V	C	C	S	D	N	Q	H	C	C	P	Q	G	F	T	C	408
CTG	GCT	CAG	GGG	TAC	TGT	CAG	AAG	GGA	GAC	ACA	ATG	GTG	GCT	GGC	CTG	1294
L	A	Q	G	Y	C	Q	K	G	D	F	M	V	A	G	L	424
GAG	AAG	ATA	CCT	GCC	CGC	CAG	ACA	ACC	CCG	CTC	CAA	ATT	GGA	GAT	ATC	1342
E	K	I	P	A	R	Q	T	T	P	L	Q	I	G	D	I	440
GGT	TGT	GAC	CAG	CAT	ACC	ATC	TGC	CCA	GTA	GGG	CAA	ACC	TGC	TGC	CCA	1390
G	C	D	Q	H	T	S	C	P	V	G	Q	T	C	C	P	456
AGC	CTC	AAG	GGA	AGT	TGG	GCC	TGC	TGC	CAG	CTG	CCC	CAT	GCT	GTG	TGC	1438
S	L	K	G	S	W	A	C	C	Q	L	P	H	A	V	C	472
TGT	GAG	GAC	CGG	CAG	CAC	TGT	TGC	CCG	GCC	GGG	TAC	ACC	TGC	AAC	GTG	1486
C	E	D	R	Q	H	C	C	P	A	G	Y	T	C	N	V	488
AAG	GCG	AGG	ACC	TGT	GAG	AAG	GAT	GTC	GAT	TTT	ATC	CAG	CCT	CCC	GTG	1534
K	A	R	T	C	E	K	D	V	D	F	I	Q	P	P	V	504
CTC	CTG	ACC	CTC	GGC	CCT	AAG	GTT	GGG	AAT	GTG	GAG	TGT	GGA	GAA	GGG	1582
L	L	T	L	G	P	K	V	G	N	V	E	C	G	E	G	520
CAT	TTC	TGC	CAT	GAT	AAC	CAG	ACC	TGT	TGT	AAA	GAC	AGT	GCA	GGA	GTC	1630
H	F	C	H	D	N	Q	T	C	C	K	D	S	A	G	V	536
TGG	GCC	TGC	TGT	CCC	TAC	CTA	AAG	GGT	GTC	TGC	TGT	AGA	GAT	GGA	CGT	1678
W	A	C	C	P	Y	L	K	G	V	C	C	R	D	G	R	552
CAC	TGT	TGC	CCC	GGT	GGC	TTC	CAC	TGT	TCA	GCC	AGG	GGA	ACC	AAG	TGT	1726
H	C	C	P	G	G	F	H	C	S	A	R	G	T	K	C	568
TTG	CGA	AAG	AAG	ATT	CCT	CGC	TGG	GAC	ATG	TTT	TTG	AGG	GAT	CCG	GTC	1774
L	R	K	K	I	P	R	W	D	M	F	L	R	D	P	V	584
CCA	AGA	CCG	CTA	CTG	TAA	GGA	AGG	GCT	ACA	GAC	TTA	AGG	AAC	TCC	ACA	1822
P	R	P	L	L	*											589
GTC	CTG	GGA	ACC	CTG	TTC	CGA	GGG	TAC	CCA	CTA	CTC	AGG	CCT	CCC	TAG	1870
CGC	CTC	CTC	CCC	TAA	CGT	CTC	CCC	GGC	CTA	CTC	ATC	CTG	AGT	CAC	CCT	1918
ATC	ACC	ATG	GGA	GGT	GGA	GCC	TCA	AAC	TAA	AAC	CTT	CTT	TTA	TGG	AAA	1966
GAA	GGC	TGT	GGC	CAA	AAG	CCC	CGT	ATC	AAA	CTG	CCA	TTT	CTT	CCG	GTT	2014
TCT	GTG	GAC	CTT	GTG	GCC	AGG	TGC	TCT	TCC	CGA	GCC	ACA	GGT	GTT	CTG	2062
TGA	GCT	TGC	TTG	TGT	GTG	TGT	GCG	CGT	GTG	CGT	GTG	TTG	CTC	CAA	TAA	2110
AGT	TTG	TAC	GCT	TTC	TGA	AAA	AAA	AAA								2137

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gccccgcgt gggacgcccc ttgagggac ccagcctga gacagctgt gtgagggaca  
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cctagcacct cccccaccc aaattctccc tggacccat tctgagctcc ccatcaccat  
gggaggtgg gccctaatct aaggcccttc cctgtcagaa ggggggtgag gcaaaagccc  
attacaagct gccatccct cccgtttca gtggacctg tggccagggt ctttcccta  
tccacagggg tgtttgtg ttgggtgtg ttcaataaa gttgtcact ttct\*

B: Amino-acid sequence of human granulin/epithelin precursor (human GP88).

MWTLVSWVALTAGLVAGTRCPDGFQFCPVACCLDPGGASYSCCRP  
LLDKWPTTLRHLGGPCQVDAHCSAGHSCIFTVSGTSSCCPFPEAVACGDGHHCCPRG  
FHCSADGRSCFQSGNNSVGAIQCPDSQFECPDFSTCCVMVDGSGWCCPMPQASCCED  
RVHCCPHGAFCDLVHTRCITPTGTHPLAKKLPAQRTNRAVALSSVMCPDARSRCPDG  
STCCELPSGKYGCCPMPNATCCSDHLHCCPQDTVCDLIQSKCLSKENATDILLTKLPA  
HTVGDVKCDMEVSCPDPGYTCRQLQSGAWGCCPFTQAVCCEDHIHCCPAGFTCDTQKGT  
CEQGPHQVPWMEKAPAHLSLPDPOALKRDVPCDNVSSCPSSDTCCQLTSGEWGCCPIP  
EAVCCSDHQHCCPQRYTCVAEGQCQRGSEIVAGLEKMPARRGSLSHPRDIGCDQHTSC  
PVGGTCCPSQGGSWACCQLPHAVCCEDRQHCCPAGYTCNVKARSCEKEVVSAQPATFL  
ARSPHVGVKDVECGEGHFCHDNQTCCRDNRQGWACCPYAQGVCCADRRHCCPAGFRCA  
RRGTKCLRREAPRWDAPLRDPALRQLL\*

FIG 9

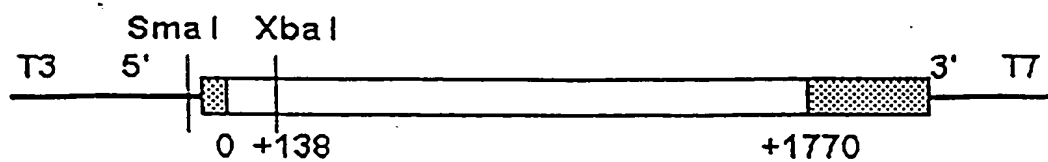
Mouse GP88 protein sequence

MVVLSVLAFAAGLVAG 17  
 TQCPDGGF--CPVA--CCLDQG--GANYCCNPLLDTPAITSHL 57  
 DQSC-QTHGHPAGY-SCLLTUSGTS--SCCPFSKGUSCGGCVHCCPQGFHCSADGKSCFQNSDNL 120  
 GAUQPGSQFECPSATCCINVD-G-SUGCCPMPQASCCEDRUHCCPHGASCDLUHTACUSPTGTHLLKKFPAQKTHRAUSLPFS 204  
 UUCPDRAKTQCPDSDICCELP-TGK-YGCCPMPHAICCSDHLHCCPQDTUCDLIQSKCLSKHYTTDLLTKLPVPUK 278  
 EVKC-DNEUSCEGYTCALN-TGA-WGCCPFKAUCCEDHHCPCPAGFOCHTEKTCENGILQUPHMKVIAAPRALPDOLLKS 360  
 DTPCQDFIR-QPTNNJCCKLN-SGD-WGCCPIPEAUCCSDNQHCCPQGFCLAQGY-CQKGDTHVAGLEKIPARQTTPLQIG 438  
 DIGCDUHT-SEPVGOTCCPSLK-G-SUACCQLPHAUCCEDRQHCCPAGYTCNUKARTCEKDUDFIQPPULLTLGPKUG 513  
 HUECGEGHF-CHDNQTCCKDSA-GU-WACCPVLKGUCCRDGRHCCPGGFHCSARGTKCLRKKIPRAUDHFLADPUPAPLL 589

consensus sequence:

C.....C.....CC.....G.....CC.....CC.D..HCCP.....C.....C

1, 2: mouse epithelin 1, 2.  
 A, B, C, D, e, f, g: granulin A, B, C, D, E, F, G; N-terminus of granulin A, B, C, D have been sequenced.  
 Mouse epithelin precursor sequence is from Plowman et al.(1992).



# Structure of pCMV<sub>4</sub> Expression Vector

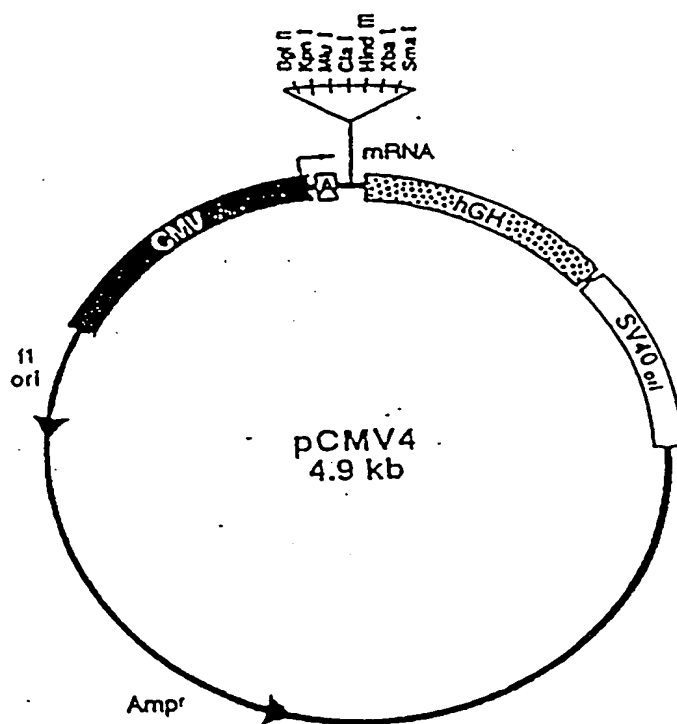


FIG. 11

FIG 12. CROSS-LINKING OF  $^{125}\text{I}$ -rGP88 TO CCL64 CELLS

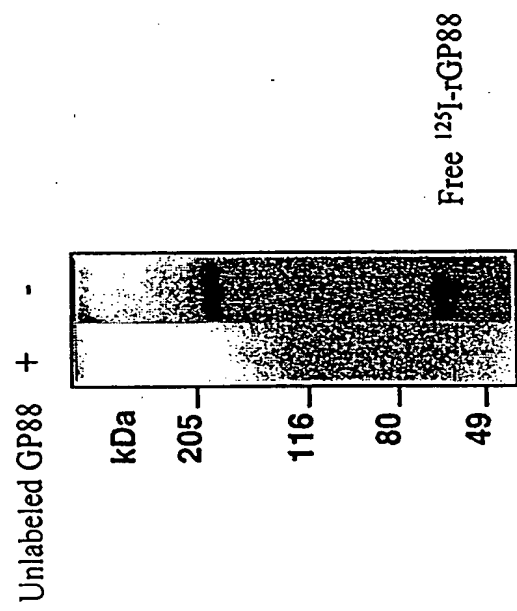
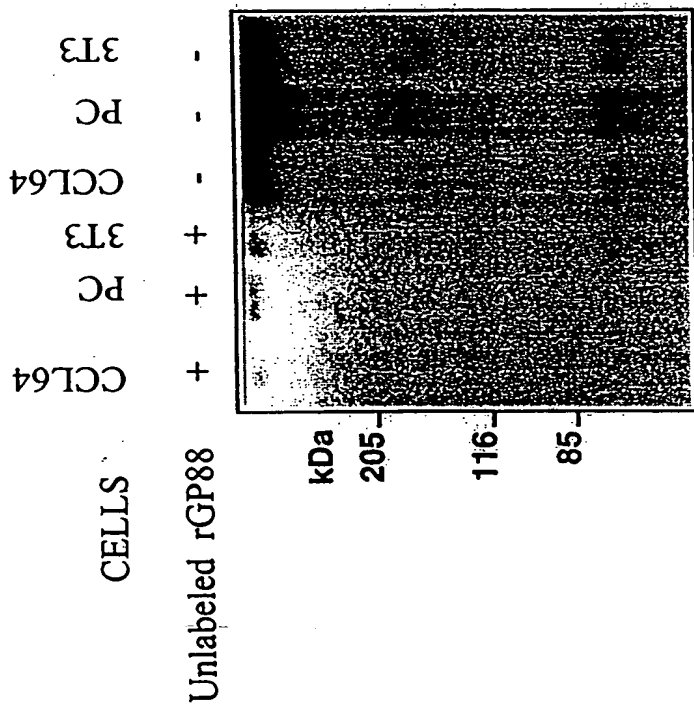
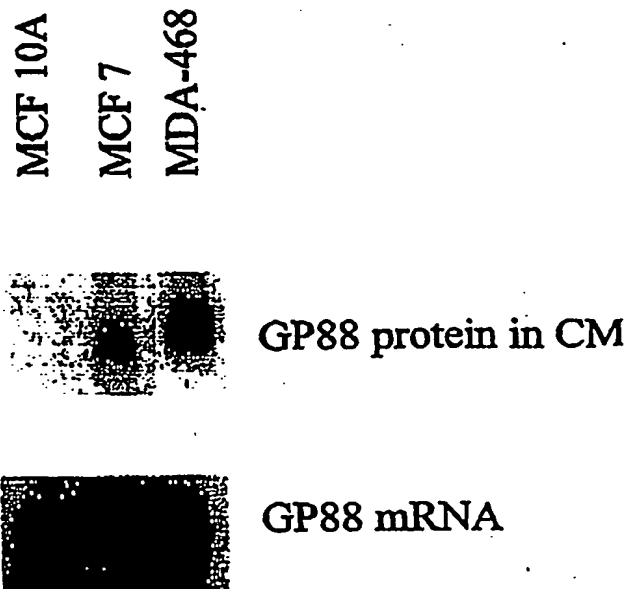


FIG 13. CROSS-LINKING OF  $^{125}\text{I}$ -rGP88





**Fig 14 GP88 Expression In Non Tumorigenic (MCF 10A) And Malignant (MCF 7, MDA-468) Human Mammary Epithelial Cells**



**Fig 15 GP88 Expression Is Inhibited By Antisense GP88 cDNA Transfection In Human Breast Carcinoma MDA-468**

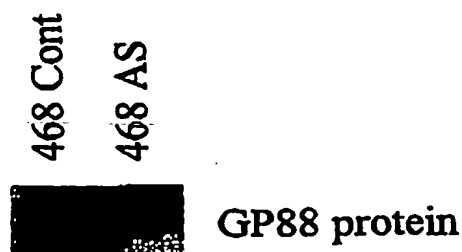


Figure 16

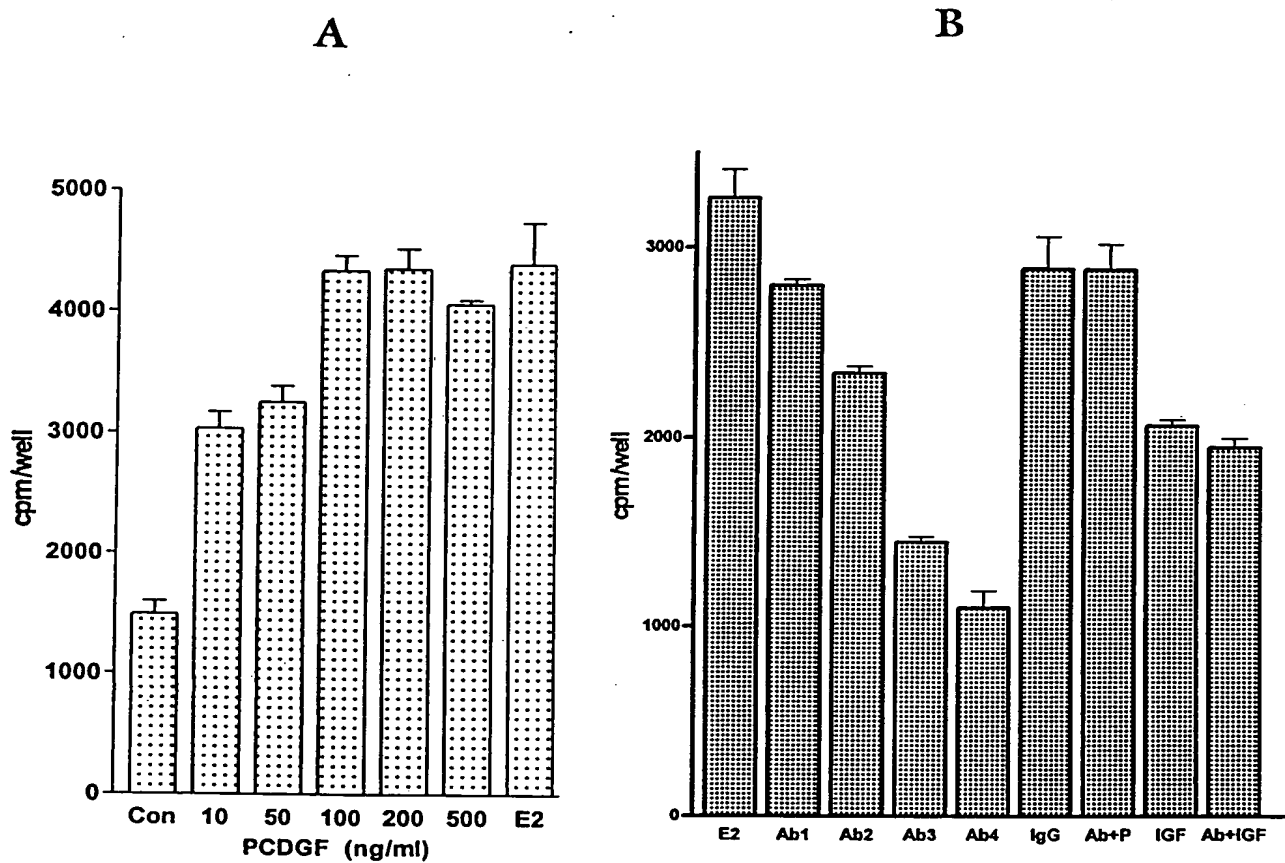


Figure 17

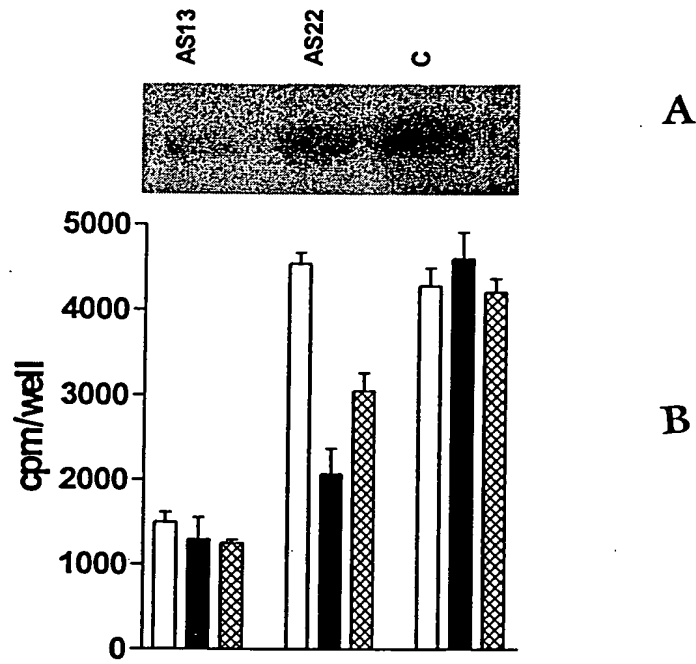


Figure 18

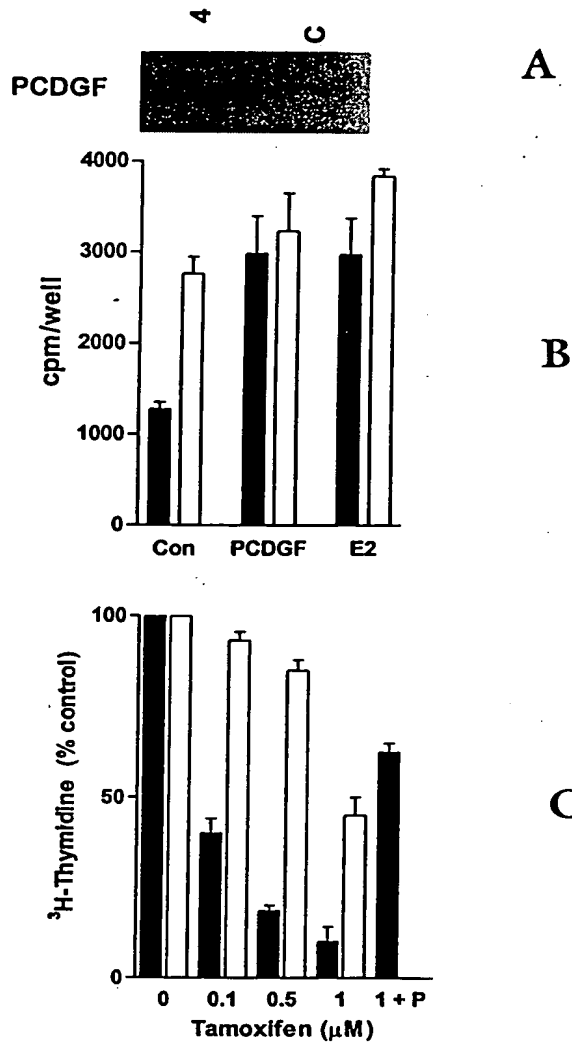
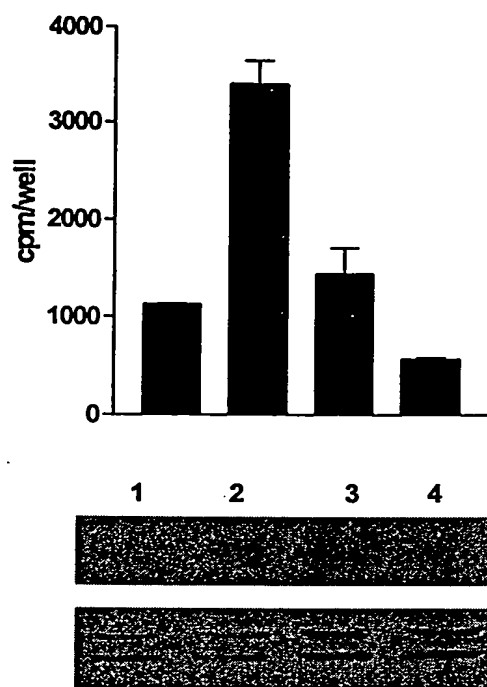


Figure 19



A

B

Figure 20

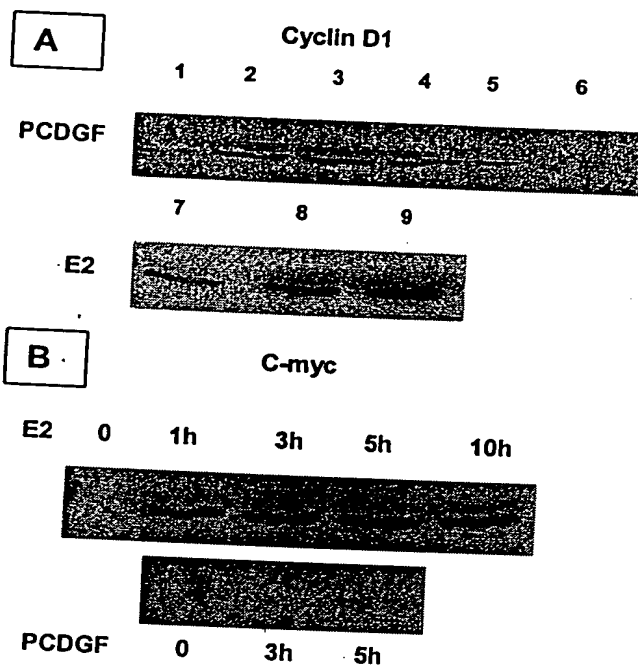
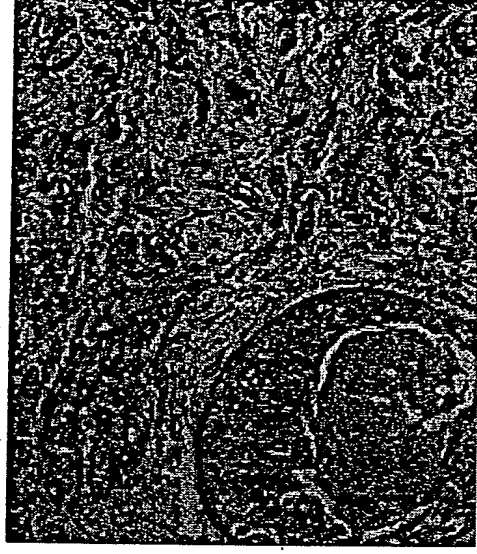


Figure 21



Benign lesion

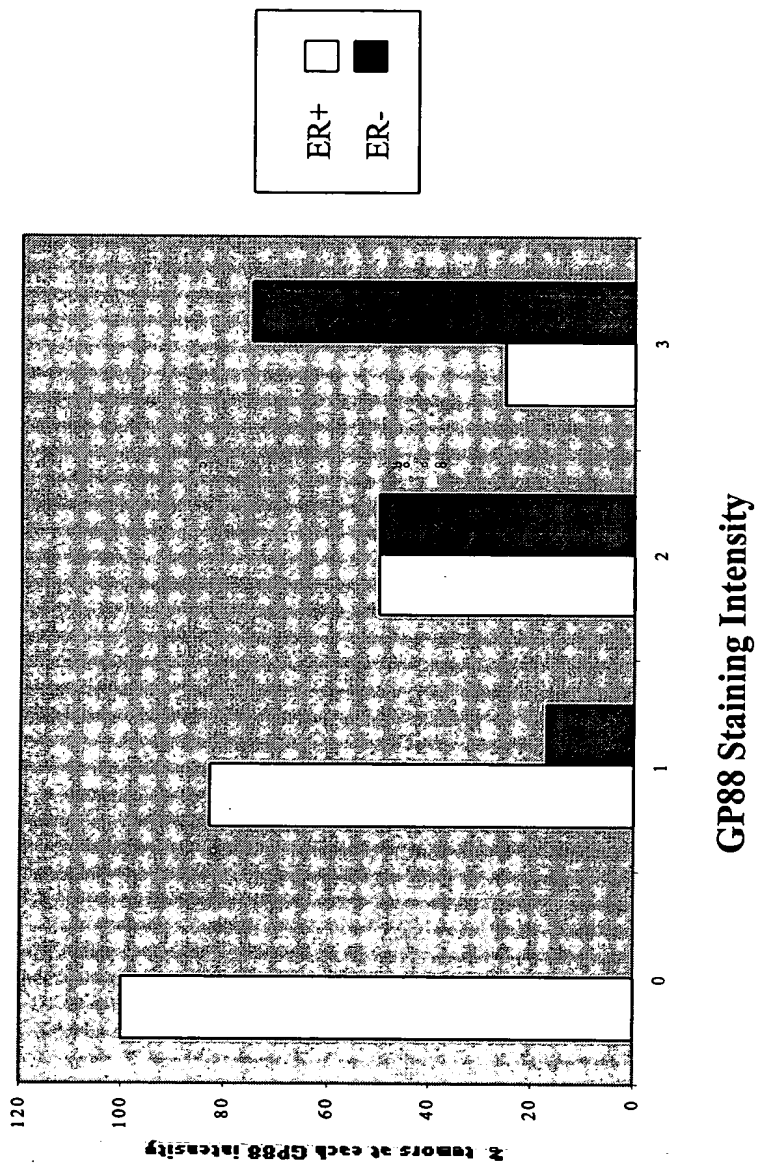


Ductal invasive Carcinoma

GP88 staining with anti-GP88 antibody  
on paraffin embedded breast cancer biopsies  
by immunohistochemistry (IHC)

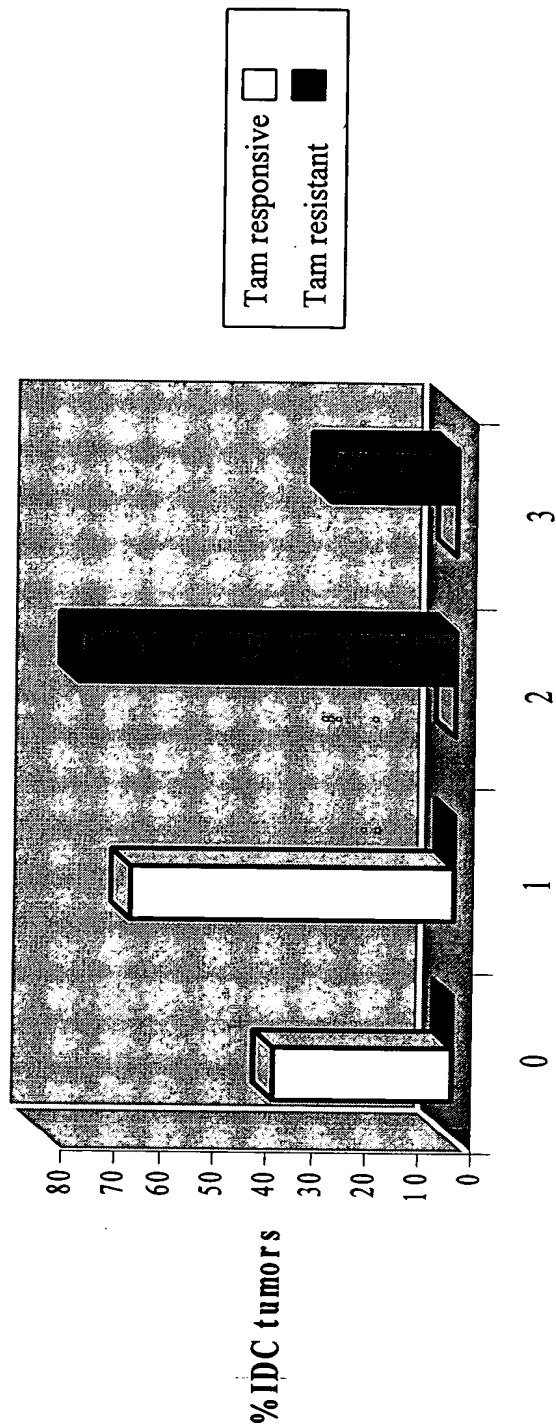
Figure 22

GP88 Staining Intensity In ER+ and ER- IDC Tumors





**Figure 23**  
**Tamoxifen responsiveness of ER+ IDC tumors classified**  
**by GP88 staining intensity**



**GP88 staining intensity**